PATENT COOPERATION TREATY

PCT

INTERNATIONAL PRELIMINARY EXAMINATION REPORT

(PCT Article 36 and Rule 70)

Applicant's or agent's file reference WJP 03 1348 8238	**FOR FURTHER ACTION**	See Notification of Transmittal of International Preliminary Examination Report (Form PCT/IPEA/416).
International Application No. **PCT/AU2003/001142**	International Filing Date *(day/month/year)* 4 September 2003	Priority Date *(day/month/year)* 4 September 2002
International Patent Classification (IPC) or national classification and IPC **Int. Cl.** [7] C12Q 1/68; C12N 15/00		
Applicant JOHNSON & JOHNSON RESEARCH PTY LTD et al		

1. This international preliminary examination report has been prepared by this International Preliminary Examining Authority and is transmitted to the applicant according to Article 36.

2. This REPORT consists of a total of 5 sheets, including this cover sheet.

   ☐ This report is also accompanied by ANNEXES, i.e., sheets of the description, claims and/or drawings which have been amended and are the basis for this report and/or sheets containing rectifications made before this Authority (see Rule 70.16 and Section 607 of the Administrative Instructions under the PCT).

   These annexes consist of a total of sheet(s).

3. This report contains indications relating to the following items:

   - I ☒ Basis of the report
   - II ☐ Priority
   - III ☐ Non-establishment of opinion with regard to novelty, inventive step and industrial applicability
   - IV ☐ Lack of unity of invention
   - V ☒ Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement
   - VI ☒ Certain documents cited
   - VII ☐ Certain defects in the international application
   - VIII ☐ Certain observations on the international application

Date of submission of the demand 30 March 2004	Date of completion of the report 21 December 2004
Name and mailing address of the IPEA/AU AUSTRALIAN PATENT OFFICE PO BOX 200, WODEN ACT 2606, AUSTRALIA E-mail address: pct@ipaustralia.gov.au Facsimile No. (02) 6285 3929	Authorized Officer **TERRY MOORE** Telephone No. (02) 6283 2632

INTERNATIONAL PRELIMINARY EXAMINATION REPORT

International application No.
PCT/AU2003/001142

I. Basis of the report

1. With regard to the elements of the international application:*

[X] the international application as originally filed.

[ ] the description, pages , as originally filed,
pages , filed with the demand,
pages , received on with the letter of

[ ] the claims, pages , as originally filed,
pages , as amended (together with any statement) under Article 19,
pages , filed with the demand,
pages , received on with the letter of

[ ] the drawings, pages , as originally filed,
pages , filed with the demand,
pages , received on with the letter of

[ ] the sequence listing part of the description:
pages , as originally filed
pages , filed with the demand
pages , received on with the letter of

2. With regard to the **language**, all the elements marked above were available or furnished to this Authority in the language in which the international application was filed, unless otherwise indicated under this item.
These elements were available or furnished to this Authority in the following language which is:

[ ] the language of a translation furnished for the purposes of international search (under Rule 23.1(b)).

[ ] the language of publication of the international application (under Rule 48.3(b)).

[ ] the language of the translation furnished for the purposes of international preliminary examination (under Rules 55.2 and/or 55.3).

3. With regard to any **nucleotide and/or amino acid sequence** disclosed in the international application, the international preliminary examination was carried out on the basis of the sequence listing:

[ ] contained in the international application in written form.

[ ] filed together with the international application in computer readable form.

[ ] furnished subsequently to this Authority in written form.

[ ] furnished subsequently to this Authority in computer readable form.

[ ] The statement that the subsequently furnished written sequence listing does not go beyond the disclosure in the international application as filed has been furnished.

[ ] The statement that the information recorded in computer readable form is identical to the written sequence listing has been furnished

4. [ ] The amendments have resulted in the cancellation of:

[ ] the description, pages

[ ] the claims, Nos.

[ ] the drawings, sheets/fig.

5. [ ] This report has been established as if (some of) the amendments had not been made, since they have been considered to go beyond the disclosure as filed, as indicated in the Supplemental Box (Rule 70.2(c)).**

* *Replacement sheets which have been furnished to the receiving Office in response to an invitation under Article 14 are referred to in this report as "originally filed" and are not annexed to this report since they do not contain amendments (Rules 70.16 and 70.17).*

** *Any replacement sheet containing such amendments must be referred to under item 1 and annexed to this report*

V. Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

1. Statement

Novelty (N)	Claims	1–24, 27, 29, 31, 34, 36, 38, 40–59	YES
	Claims	25, 26, 28, 30, 32, 33, 35, 37, 39	NO
Inventive step (IS)	Claims	1–24, 40–59	YES
	Claims	25–39	NO
Industrial applicability (IA)	Claims	1-59	YES
	Claims		NO

2. Citations and explanations (Rule 70.7)

The applicant's invention resides in the provision of methods of producing dsDNA molecules that can be used to mediate RNA interference (RNAi). The claims relate to different aspects of this principle, as follows.

Claims 1–7 and 13–19 relate to methods of DNA production that provide a hairpin downstream of a random sequence, in which the hairpin is used to prime synthesis of a complementary DNA, which is then denatured, hybridised to a primer and used to synthesise dsDNA.

Claims 8–12, 20–24 and 48–55 relate to methods of DNA production in which a library of DNA fragments is adapted to provide at least four consecutive adenosine nucleotides at the 3' end, and is then cloned between two convergent promoters.

Claims 25–39 relate to expression vectors comprising DNA cloned between convergent promoters and transcription terminators.

Claims 40–47 and 56–59 relate to methods of producing DNA libraries based on ligating hairpin DNA or RNA to a library of dsDNA, followed by denaturation, priming and extension to produce a library of dsDNA molecules.

The following documents identified in the International Search Report have been considered for the purposes of this report.

D1:Morris JC et al (2 September 2002).
D2: Wang Z et al (2000).
D3: EP 1 229 134 (A2)

Novelty (N)

D1 discloses an RNAi library generated using the pZJMβ vector. This vector has convergent T7 promoters and a multiple cloning site flanked by two directional transcription terminators. This expression system was used for forward genetic screening of gene function in *Trypanosoma brucei*. As such, the citation is novelty-destroying for claims 25, 26, 28, 30, 32, 33, 35, 37 and 39.

D2 discloses the construction and use of an RNAi expression vector, pZJM. This vector has convergent T7 promoters and a cloning site flanked by two directional transcription terminators. As such, the citation is novelty-destroying for claims 25, 26, 28, 30 and 39.

None of the citations anticipate the features of claims 1–24, 27, 29, 31, 34, 36, 38 and 40–59. Therefore, these claims are considered to be novel.

(continued on supplemental sheet)

VI. Certain documents cited

1. Certain published documents (Rule 70.10)

Application No. Patent No.	Publication date *(day/month/year)*	Filing date *(day/month/year)*	Priority date (valid claim) *(day/month/year)*
WO 03/046186	05 June 2003	28 November 2002	28 November 2001
WO 03/046173	05 June 2003	30 October 2002	28 November 2001

These documents disclose *in vivo* siRNA expression systems comprising 19 base pair DNA fragments flanked by four consecutive adenosine nucleotides and four consecutive thymidine nucleotides (see abstracts and Figures 1/32).

With regard to the document(s) listed in Box VI under "certain documents cited", these are documents published prior to the international filing date but later than the priority date claimed but which would otherwise be considered to be of particular relevance.

Under the PCT, novelty is considered only in respect of documents published before the priority date. The relevance of a document published after the priority date is dependent upon national law. Such documents are excluded from consideration in preliminary examination, under the PCT Guidelines but have been included here for information.

2. Non-written disclosures (Rule 70.9)

Kind of non-written disclosure	Date of non-written disclosure *(day/month/year)*	Date of written disclosure referring to non-written disclosure *(day/month/year)*

Supplemental Box
(To be used when the space in any of the preceding boxes is not sufficient)

Continuation of Box V

Inventive step (IS)

The disclosures of D1 and D2 are discussed above under Novelty. Claims 29, 31, 36 and 38 differ from D1, and claims 29, 31–33 and 35–38 differ from D2 in the choice of standard expression vector components used. These features are technical equivalents of those of the citations. Therefore, these claims lack inventive step in the light of either D1 or D2.

Claims 27 and 34 recite RNAi sequences of 19 base pairs in length. Neither D1 nor D2 suggests such short fragments, and therefore these claims are considered to be inventive in the light of these citations.

D3 discloses the construction of RNAi expression vectors using convergent promoters (see paragraph [0113]). These RNAi constructs include short nucleotide fragments (see, for example, paragraph [0015]). The citation does not, however, disclose the use of two directional transcription terminators as recited by claims 25–39. However, the use of such terminators is considered to be a standard choice of components used in expression vectors. This feature is a technical equivalent of the citation. Therefore, these claims lack inventive step in the light of D3.

Claims 1–24 and 40–59 recite features that are considered inventive in the light of the citations.

Industrial applicability (IA)

Claims 1–59 meet the requirements of the PCT with regard to industrial applicability.

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1. Statement

Novelty (N)	Claims	**1–24, 27, 29, 31, 34, 36, 38, 40–59**	**YES**
	Claims	**25, 26, 28, 30, 32, 33, 35, 37, 39**	**NO**
Inventive step (IS)	Claims	**1–24, 40–59**	**YES**
	Claims	**25–39**	**NO**
Industrial applicability (IA)	Claims	**1-59**	**YES**
	Claims		**NO**

2. Citations and explanations (Rule 70.7)

The applicant's invention resides in the provision of methods of producing dsDNA molecules that can be used to mediate RNA interference (RNAi). The claims relate to different aspects of this principle, as follows.

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